

# Myeloid-derived suppressor cells in allogeneic hematopoietic stem cell transplantation

## A double-edged sword?

Katarina Le Blanc<sup>1</sup>, Regina Jitschin<sup>1,2</sup>, and Dimitrios Mougakakos<sup>2,\*</sup>

<sup>1</sup>Department of Medicine; Karolinska Institutet; Hematology Center; Karolinska University Hospital; Stockholm, Sweden;

<sup>2</sup>Department of Hematology and Oncology; University Hospital Erlangen-Nuremberg; Erlangen, Germany

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**Abbreviations:** allo-HSCT, allogeneic hematopoietic stem cell transplantation; GvHD, graft- vs. -host disease; GvT, graft- vs. -tumor; IDO, indoleamine-2,3-dioxygenase; MDSC, myeloid derived suppressor cell

Accumulating evidence suggests that myeloid-derived suppressor cells (MDSCs) underpin an immunological checkpoint that is activated during inflammation or "inflammatory-like" conditions like cancer. Here, we discuss the identification of MDSCs in patients receiving allogeneic hematopoietic stem cell transplantation and their potential as therapeutic targets or tools for improving the efficacy of this treatment.

Nowadays, allogeneic hematopoietic stem cell transplantation (allo-HSCT) represents the only curative regimen for a number of malignancies. The therapeutic success of allo-HSCT based on the immunological interaction between donor and recipient cells, highlighting the ability of the immune system to eradicate malignant lesions, at least under selected circumstances. The most prominent cell populations implicated in this process, which is commonly known as graft- vs. -tumor (GvT) effect, are T and natural killer (NK) cells. However, such a beneficial effect—which results from the immunological non-identity between the host and the donor—is regularly linked to an unwarranted involvement of healthy recipient tissues. The so-called graft- vs. -host disease (GvHD) is largely responsible for transplantation-related mortality and morbidity.<sup>1</sup> During the process of immune reconstitution that follows allo-HSCT, a delicate immunological balance determines whether patients develop a therapeutic GvT effect or a self-limiting GvHD, and at the same time whether they successfully withstand

microbial challenges. Thus, deciphering the immune components that orchestrate GvT reactions as opposed to GvHD has attracted major interest.

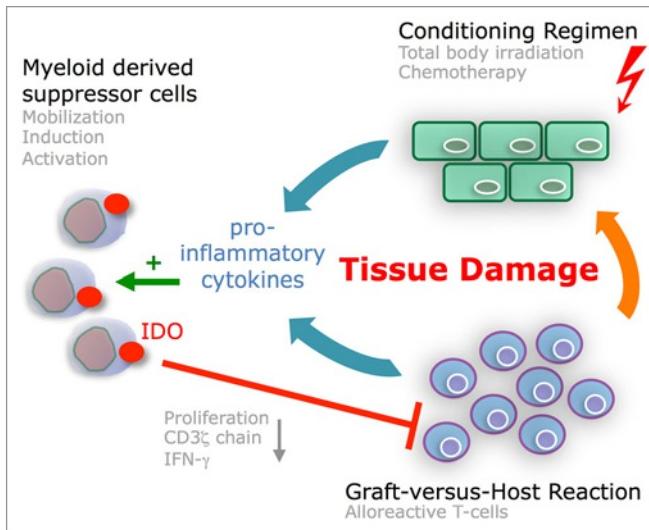
Myeloid-derived suppressor cells (MDSCs) have been recently recognized as potential players in the setting of allo-HSCT. During the past decade, MDSCs have been intensively investigated as part of the immune cells that infiltrate solid tumors such as renal cancer and melanoma.<sup>2</sup> MDSCs have been shown to exert robust immunosuppressive functions, thus representing the myeloid counterpart of regulatory T cells (Tregs). The accumulation of MDSCs in cancer patients is thought to stem from an increased mobilization of myeloid precursors from the bone marrow, a differentiation block and a re-programming of matured myeloid cells toward an immature and immunosuppressive phenotype. Most likely, all these mechanisms act simultaneously in cancer patients, explaining the vast heterogeneity of MDSCs. Despite some extent of heterogeneity (in terms of phenotype, immunosuppressive mechanisms and defects in signaling pathways associated

with myeloid differentiation), which presumably originates from disease-specific shaping microenvironments, a relatively defined dichotomy has been proposed to classify MDSCs in a monocytic and a granulocytic subset. The MDSC repertoire of T and NK cell-suppressive mechanisms is very broad and includes the secretion of cytokines (e.g., transforming growth factor  $\beta$ , TGF $\beta$ ), the production of reactive oxygen species, and the depletion of vital amino acids (e.g., arginine). One common denominator of the accumulation of MDSCs is a driving inflammatory milieu. Cytokines that are abundant during inflammation such as tumor necrosis factor  $\alpha$  (TNF $\alpha$ ), granulocyte colony-stimulating factor (G-CSF), and interleukin (IL)-6 are linked to MDSC levels in patients and stimulate the accumulation of MDSCs in vitro.<sup>3</sup> Since increased MDSC levels are not limited to neoplastic entities but occur in the course of various inflammatory conditions, it is tempting to speculate that MDSCs underpin an intrinsic checkpoint for the control of immune responses. Such a checkpoint turns against the host during cancer, as

\*Correspondence to: Dimitrios Mougakakos; Email: dimitrios.mougakakos@uk-erlangen.de

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**Figure 1.** Accumulation of myeloid-derived suppressor cells in allo-HSCT. Preparatory (radio-) chemotherapy (conditioning regimen) for allogeneic hematopoietic stem cell transplantation (allo-HSCT) leads to tissue damage and subsequent inflammation. In addition, alloreactive donor T cells—which are fundamental for establishing the graft- vs. -tumor (GvT) effect—regularly target healthy tissue (preferably the liver, skin, and gut) of the host, resulting in a highly inflammatory condition known as graft- vs. -host disease (GvHD). The high levels of pro-inflammatory cytokines released in these setting stimulate myeloid-derived suppressor cells (MDSCs) as part of a physiological mechanism for the control of inflammatory damage. This stimulation most likely represents a multistep process, including an increased mobilization of myeloid progenitors from the bone marrow, a differentiation arrest and the re-programming of mature myeloid cells mediate immunosuppressive functions. Accumulating monocytic MDSCs following allo-HSCT express the enzyme indoleamine-2,3-dioxygenase (IDO), which exerts suppressive effects on T cells by depleting tryptophan and via its metabolites (e.g., kynurenine). Such immunosuppressive effects include the inhibition of T-cell proliferation and interferon  $\gamma$  (IFN $\gamma$ ) production as well as the downregulation of CD3 $\zeta$ .

the latter often represents an “inflammatory-like” condition.

Interestingly, several factors that might promote the accumulation of MDSCs come together in the course of allo-HSCT: (1) the preparatory (radio-)chemotherapeutic regimen (best known as conditioning) causes tissue damage and inflammation, (2) regenerative myelopoiesis favors the efflux of immature cell progenitors from the bone marrow during early engraftment, and (3) cytopenia results in a compensatory release of various cytokines including myelopoietic growth factors. In fact, a relative moncytosis is frequently observed in transplanted patients, regularly lasting for a year after allo-HSCT. We found an increased proportion of HLA-DR<sup>low</sup> or HLA-DR<sup>neg</sup> cells among these monocytes, resulting in an overall increase in the frequency of CD14<sup>+</sup>HLA-DR<sup>low/neg</sup> cells as compared with the healthy individuals.<sup>4</sup> This surface phenotype is typical of the monocytic MDSCs that are found within various neoplastic lesions, including melanoma.<sup>5</sup> Of note, describing an aberrant myeloid phenotype is not sufficient for defining MDSCs. Rather, an experimental proof

of their immunosuppressive functions is obligatory. Therefore we purified the cells of interest and performed immunosuppression assays using activated autologous T cells. We observed a dose-dependent suppression of T cells, allowing us to term the myeloid cells we purified MDSCs. Cells with the same surface phenotype isolated from healthy subjects also exerted an immunosuppressive activity, albeit to a substantially lower extent. Together with the recent discovery that naturally occurring CD14<sup>+</sup>HLA-DR<sup>low/neg</sup> cells can stimulate Tregs,<sup>6</sup> our findings corroborate the notion that MDSCs underpin an immunological checkpoint that also operates in physiological conditions.

In line with the aforementioned critical connection between cytokines and MDSCs, we found a significant correlation between the levels of MDSCs and the concentration of G-CSF, IL-6, and IL-10 in the patients’ sera. These cytokines as well as MDSCs declined steadily over time following a peak that occurs early after allo-HSCT, a moment that is often characterized by a so-called “cytokine storm.” Our mechanistic analyses revealed indoleamine-2,3-dioxygenase (IDO) as

the key mediator of the immunosuppressive activity of MDSCs in allo-HSCT. This enzyme, which catalyzes the rate-limiting step of tryptophan degradation, has a well-known tolerogenic activity, plays a major role in fetal tolerance and is often linked to tumor-induced immunosuppression. Blocking IDO in MDSCs increased T-cell proliferation, stimulated interferon  $\gamma$  (IFN $\gamma$ ) production, and reduced the incidence of T-cell apoptosis. Interestingly, patients with severe GvHD had high levels of MDSCs, which is in line with a generalized activation of tryptophan metabolism during GvHD.<sup>7</sup> This finding could be indicative of a compensatory increase in regulatory myeloid cell subsets, which however fail to control the overwhelming immune response that underpins GvHD.

Based on these findings, it is tempting to speculate that—similar to Treg-based strategies—MDSC-based approaches might constitute a therapeutic option for GvHD. Murine models have generate promising data in this sense, demonstrating that the adoptive transfer of MDSCs can result in the successful control of GvHD without compromising GvT effects.<sup>8,9</sup> In addition,

the production of clinical-grade monocytic MDSCs using cytokine cocktails appears feasible. However, studies evaluating large patient cohorts for long observational periods are required to elucidate the actual potential of MDSCs in patients receiving allo-HSCT. First, we need to unequivocally clarify the impact of MDSCs on the

risk of disease. Second, we have to carefully assess whether MDSCs are linked to viral infection and/or reactivation, since an increasing number of publication suggests that viruses (e.g., hepatitis C virus) utilize MDSCs for escaping immunosurveillance.<sup>10</sup> Irrespective of these incognita, MDSCs are getting under the limelight on

the allo-HSCT stage and might represent a promising therapeutic tool for the management of GvHD or target for improving the GvT effect (Fig. 1).

#### Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

#### References

1. Ferrara JL, Levine JE, Reddy P, Holler E. Graft-versus-host disease. *Lancet* 2009; 373:1550-61; PMID:19282026; [http://dx.doi.org/10.1016/S0140-6736\(09\)60237-3](http://dx.doi.org/10.1016/S0140-6736(09)60237-3)
2. Gabrilovich DI, Ostrand-Rosenberg S, Bronte V. Coordinated regulation of myeloid cells by tumours. *Nat Rev Immunol* 2012; 12:253-68; PMID:22437938; <http://dx.doi.org/10.1038/nri3175>
3. Lechner MG, Liebertz DJ, Epstein AL. Characterization of cytokine-induced myeloid-derived suppressor cells from normal human peripheral blood mononuclear cells. *J Immunol* 2010; 185:2273-84; PMID:20644162; <http://dx.doi.org/10.4049/jimmunol.1000901>
4. Mougiakakos D, Jitschin R, von Bahr L, Poschke I, Gary R, Sundberg B, et al. Immunosuppressive CD14+HLA-DRlow/neg IDO+ myeloid cells in patients following allogeneic hematopoietic stem cell transplantation. *Leukemia* 2013; 27:377-88; PMID:22828446; <http://dx.doi.org/10.1038/leu.2012.215>
5. Poschke I, Mougiakakos D, Hansson J, Masucci GV, Kiessling R. Immature immunosuppressive CD14+HLA-DR-/low cells in melanoma patients are Stat3hi and overexpress CD80, CD83, and DC-sign. *Cancer Res* 2010; 70:4335-45; PMID:20484028; <http://dx.doi.org/10.1158/0008-5472.CAN-09-3767>
6. Hoechst B, Gamrekelashvili J, Manns MP, Greten TF, Korangy F. Plasticity of human Th17 cells and iTregs is orchestrated by different subsets of myeloid cells. *Blood* 2011; 117:6532-41; PMID:21493801; <http://dx.doi.org/10.1182/blood-2010-11-317321>
7. Landfried K, Zhu W, Waldhier MC, Schulz U, Ammer J, Holler B, et al. Tryptophan catabolism is associated with acute GVHD after human allogeneic stem cell transplantation and indicates activation of indoleamine-2,3-dioxygenase. *Blood* 2011; 118:6971-4; PMID:22039265; <http://dx.doi.org/10.1182/blood-2011-06-357814>
8. Highfill SL, Rodriguez PC, Zhou Q, Goetz CA, Koehn BH, Veenstra R, et al. Bone marrow myeloid-derived suppressor cells (MDSCs) inhibit graft-versus-host disease (GVHD) via an arginase-1-dependent mechanism that is up-regulated by interleukin-13. *Blood* 2010; 116:5738-47; PMID:20807889; <http://dx.doi.org/10.1182/blood-2010-06-287839>
9. Wang D, Yu Y, Haarberg K, Fu J, Kaoaard K, Nagaraj S, et al. Dynamic change and impact of myeloid-derived suppressor cells in allogeneic bone marrow transplantation in mice. *Biol Blood Marrow Transplant* 2013; 19:692-702; PMID:23376089; <http://dx.doi.org/10.1016/j.bbmt.2013.01.008>
10. Tacke RS, Lee HC, Goh C, Courtney J, Polyak SJ, Rosen HR, et al. Myeloid suppressor cells induced by hepatitis C virus suppress T-cell responses through the production of reactive oxygen species. *Hepatology* 2012; 55:343-53; PMID:21953144; <http://dx.doi.org/10.1002/hep.24700>